Structure

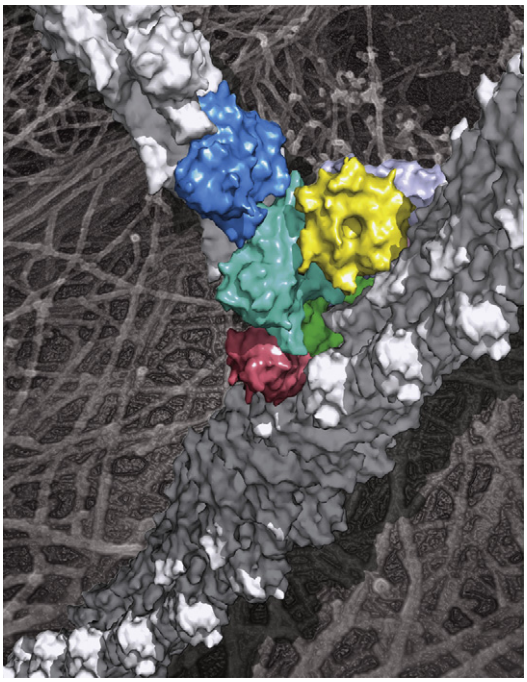
In This Issue

The Way NFAT Recognizes HIV-1 LTR DNA Elements

PAGE 684

Host factor NFAT regulates the transcription and replication of HIV-1 by binding to a tandem of two κB sites on the HIV-1 LTR. The structural and biochemical analyses reported by Bates et al. reveal surprising findings on how NFAT recognizes the two identical κB elements on the HIV-1 LTR in distinct modes and how context-dependent DNA recognition by NFAT is affected by neighboring factors and DNA conformation. These studies have general implications for understanding the combinatorial mechanism of eukaryotic gene regulation and how this mechanism may be adopted by viruses to hijack host transcription machineries for their replication.

Arp2/3 Activated Complex Story



PAGE 695

Arp2/3 complex plays a critical role in actin filament nucleation and branching during processes such as cell migration and endocytosis. Previous structures of Arp2/3 complex reveal its inactive conformation. The study of the activated complex has been hampered by uncontrollable polymerization. Boczkowska et al. engineered a stable activated nine protein complex, including the activator region of N-WASP and one actin monomer, and studied its structure in solution by X-ray scattering. Their results support a model of Arp2/3 complex activation that is consistent with most of the biochemical observations. (Figure by Boczkowska et al.)

Leucine-Rich Repeat Domain Folding Needs a Cap

PAGE 705

Courtemanche and Barrick characterize the transition state ensemble of the leucine-rich repeat (LRR) domain of Internalin B (InlB). The authors find that InlB folds via a discrete transition state in which the first three repeats and N-terminal helical cap are well structured, but the C-terminal repeats are not. Folding by a discrete transition state is surprising given the opportunity for parallel pathways, supporting the idea that subtle factors are key for pathway selection. The authors suggest that the helical caps may serve as general nuclei for folding, thus proposing a role for this structural motif commonly found in LRR proteins.

Seeing Full-Length p97 More Clearly

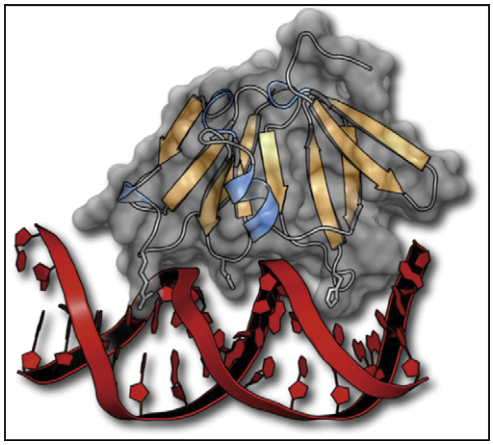
PAGE 715

Proteins of the AAA+ superfamily function in ATP-driven unfolding, dissociation, and remodeling of macromolecular complexes. The protein p97 belongs to a subgroup of AAA+ proteins that contains two nucleotide-binding domains. Interpretation of low-resolution diffraction data from crystals of full-length p97 has been hampered by the lack of a high-resolution structure of the active ATPase domain D2. Davies et al. determined the crystal structure of D2 and used it to improve the refinement of full-length p97 in different nucleotide states. The improved structures suggest pathways for transforming nucleotide hydrolysis into conformational changes.

AgrA Binding to DNA Leads to Serious Bending

PAGE 727

Sidote et al. report the crystal structure of the DNA binding domain of *Staphylococcus aureus*, AgrA, bound to DNA. This represents the first structure of a LytTR domain, a DNA binding motif found within the AlgR/AgrA/LytR family of transcription factors that regulate expression of virulence genes in pathogenic bacteria. The structure establishes a unique β fold with a mode of interaction with DNA not previously described: loop regions of AgrA contact two successive major grooves and the intervening minor groove inducing a substantial bend in the DNA. (Figure by Sidote et al.)



How Does Calmodulin Bind?

PAGE 736

Calmodulin is a ubiquitous protein that plays a key role in calcium-mediated signal transduction. Gsponer et al. apply combination of NMR spectroscopy and computational methods and demonstrate that the Ca^{2+} bound state of calmodulin includes a range of structures similar to those present in the complex with myosin light chain kinase. By bringing together modern free energy landscape theory with classical allostery models, the authors suggest that a coupled equilibrium shift mechanism controls the efficient binding of calmodulin to a wide range of ligands.

When Scorpio Toxin Attacks

PAGE 747

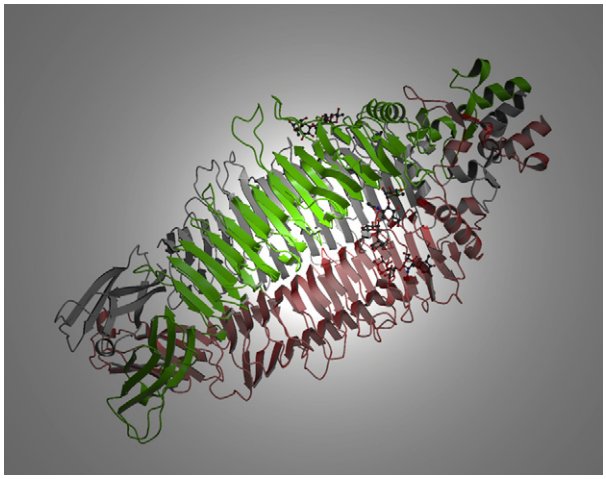
An atomistic view of the binding of a scorpio toxin to a potassium channel is presented by Zachariae et al. The tight complex was found to form spontaneously in molecular dynamics simulations and shows conformational changes in the selectivity filter of the channel that are related to the recovery from C-type inactivation. Furthermore, a binding heterogeneity was observed that may entropically stabilize the complex. Solid-state NMR and electrophysiological experiments corroborated the MD simulation findings.

Hsp90: It's All about the Change

PAGE 755

The molecular chaperone Hsp90 is required for the activation of a wide range of signaling and regulatory proteins. Hsp90's chaperone function depends upon its intrinsic ATPase activity, which drives functionally relevant conformational changes. Using small angle X-ray scattering and recently developed modeling methods, Krukenberg et al. identified a conformation of the Hsp90 bacterial homolog, HtpG, not previously described. Interestingly, AMPPNP binding incompletely converts the open apo form into the closed ATP state, and multiple conformations coexist in equilibrium. The study provides a view of Hsp90 conformational dynamics and the role of nucleotide in effecting conformational change.

Sf6 Tailspike Protein Active Site Different from Others



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The bacteriophage Sf6 infects *Shigella* by insertion of its double-stranded DNA after attaching to the host cell specifically via its tailspike proteins (TSP). The crystal structure of Sf6 TSP, reported by Müller et al., reveals a conserved trimeric architecture with a central right-handed β helix. The C-terminal domain consists of a β sandwich reminiscent of viral capsid proteins. An endorhamnosidase active site is located between two β helix subunits each anchoring one catalytic carboxylate. The functionally and structurally related bacteriophage P22 TSP has its active sites on single subunits. Sf6 TSP may thus serve as an example for the evolution of different host specificities on a similar general architecture. (Figure by Müller et al.)

α Helices Dominate Partitivirus Structure

PAGE 776

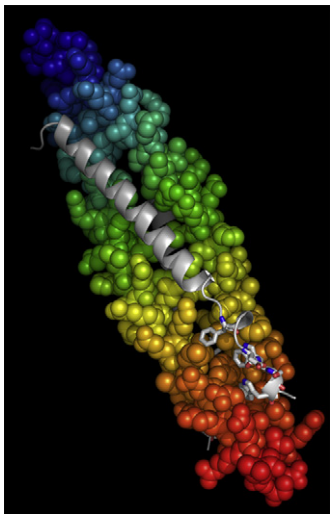
Partitiviruses are small, bisegmented dsRNA viruses that infect fungi and plants. Their two genome segments are separately packaged in icosahedral capsids containing 120 coat subunits. Ochoa et al. now describe the structure of a partitivirus, *Penicillium stoloniferum* virus S, obtained using transmission electron cryomicroscopy and 3D image reconstruction to a resolution of 7.3 Å. The structure is dominated by α helices and notable for prominent surface arches formed by quasisymmetric dimers of coat protein. This organization of the 120 coat subunits is distinct from that described for other such "T = 2" viruses that have been studied to date.

α Helices in a Membrane More Rigid More Uniform

PAGE 787

Membrane protein structure and dynamics are significantly influenced by the lipid bilayer through a range of factors. The combination of these environmental properties and the protein's amino acid composition, which is highly hydrophobic (i.e., chemically inert), result in a shifting of the balance of forces that stabilize protein structure. Page et al. now find that transmembrane α helices frequently exhibit rigidity and highly uniform structures with a conformation that is more like that of the original Pauling and Corey (1950) model than that found in water soluble proteins.

Structure-Based Explorations of NEMO



PAGE 798

NEMO is required to bind the IKK kinases and activate the NF-κB pathway leading to inflammation and uncontrolled cell growth. Previously, a hexapeptide derived from the IKK kinase was shown to block this molecular interaction and inhibited NF-κB activation. Through biophysical measurements and crystallography, Rushe et al. now describe the dimeric NEMO complexed with two IKK C termini. The work explains the role of serine phosphorylation in disruption of the complex structure, suggests that NEMO is ordered by IKK, and identifies residues beyond the hexapeptide to be mimicked by a drug. (Figure by Rushe et al.)

Mysterious SapC Domain-Swapped Dimer Finally Here

PAGE 809

Saposins are involved in membrane sphingolipid degradation and facilitate the extraction of antigenic lipids from intralysosomal membranes for loading to CD1b molecules, which present them to T cells. Here, Rossmann et al. report the crystal structure of SapD and discuss its mode of interaction with membranes containing phospho- or sulfolipids. Additionally, the authors present the crystal structure of a thus far unknown domain-swapped dimer of SapC, whose existence has been previously questioned, although it was postulated to account for the potential of SapC to promote membrane fusion.

Choosing the Right BH3 Domain for the Job

PAGE 818

Interaction of the proapoptotic BH3-only proteins with prosurvival Bcl-2 proteins triggers cell death. Some BH3-only proteins bind to all prosurvival proteins while others only interact with specific partners. Now, structural analysis of the prosurvival protein A1 bound to the BH3 domains from four different proapoptotic proteins reveals a remarkably plastic binding groove on A1 that can accommodate variable sequences (Smits et al.). Comparison of the A1 complex structures suggests that certain conserved interactions are required for optimal binding and highlights features that can be exploited for the development of molecules that selectively bind to specific prosurvival proteins.

